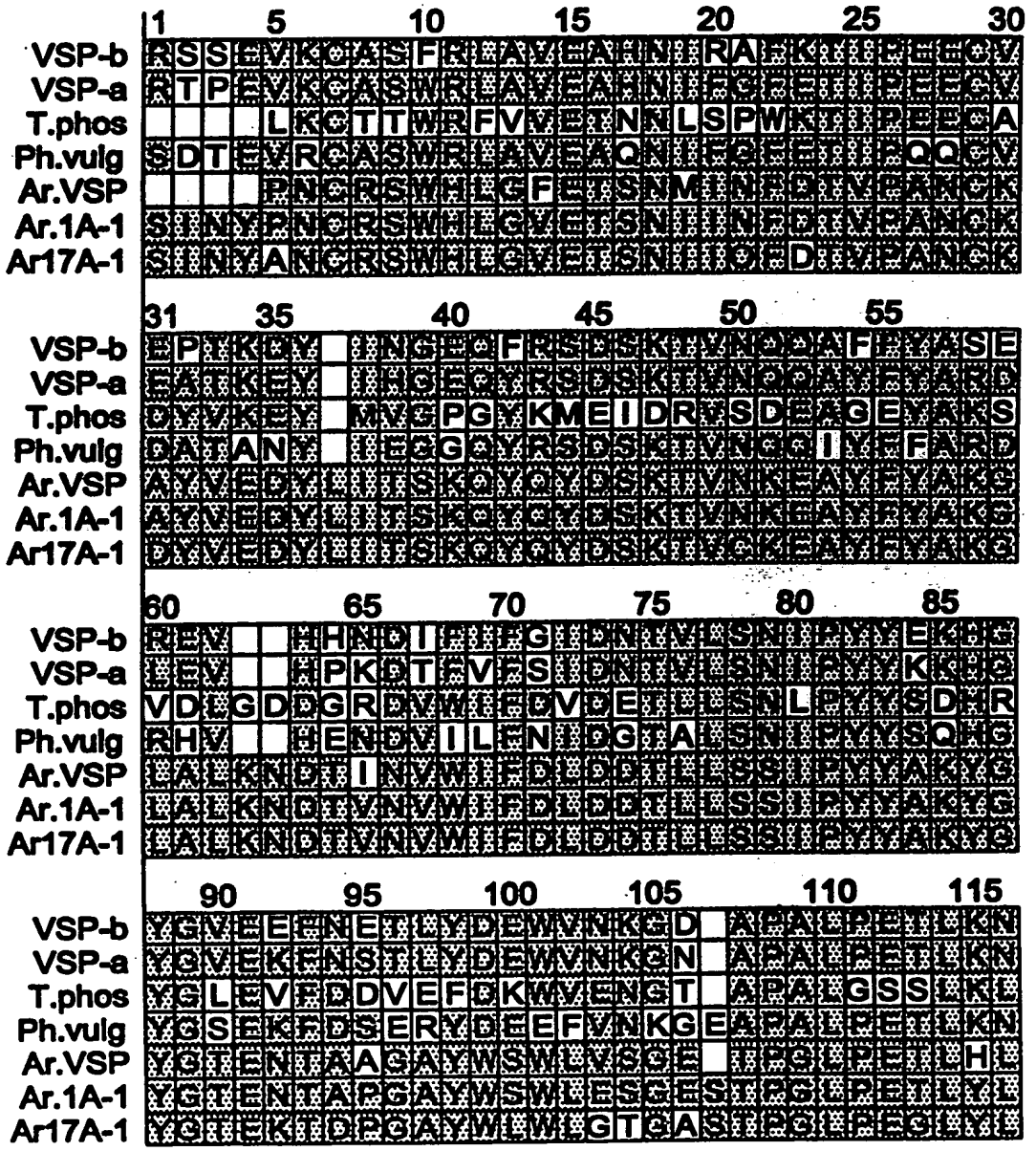
VSP HOMOLOGIES



TO FIG. 1A.

FIG. 1.

FROM FIG. 1.

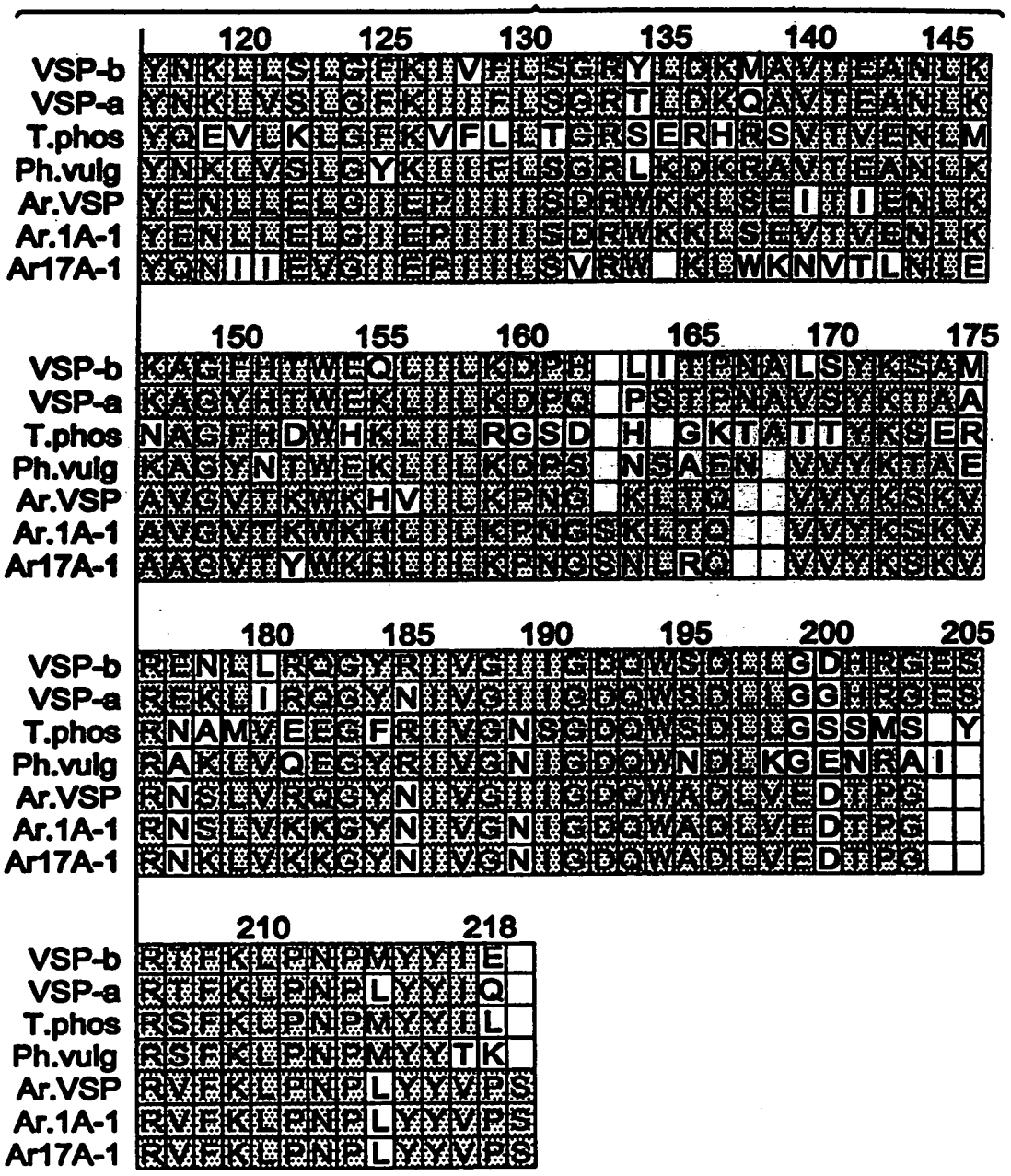


FIG. 1A.

PROPOSED VSPβ METHIONINE-ENRICHED VARIANTS

	1				5				10				15				20				25				30
VSPβ	R	S	S	E	V	K	C	A	S	F	R	L	A	V	E	A	H	N	I	R	A	F	K	T	I	P	E	E	C	V
VSPβ-Met10					M														M											M
VSPβ-Met20					M														M											M
VSPβ-Met30					M														M						M					M

	31				35				40				45				50				55				60
VSPβ	E	P	T	K	D	Y	I	N	G	E	Q	F	R	S	D	S	K	T	D	N	Q	Q	A	F	F	Y	A	S	E	R
VSPβ-Met10							M						M																	M
VSPβ-Met20	M						M						M																	M
VSPβ-Met30	M						M						M						M											M

	61				65				70				75				80				85				90
VSPβ	E	V	H	H	N	D	I	F	I	F	G	I	D	N	T	V	L	S	N	I	P	Y	Y	E	K	H	G	Y	G	V
VSPβ-Met10	M				M				M				M																	
VSPβ-Met20	M		M		M				M				M																	M
VSPβ-Met30	M		M		M				M				M				M													M

	91				95				100				105				110				115				120
VSPβ	E	E	F	N	E	T	L	Y	D	E	W	V	N	K	G	D	A	P	A	L	P	E	T	L	K	N	Y	N	K	L
VSPβ-Met10																														
VSPβ-Met20							M				M																			
VSPβ-Met30		M					M				M																			

	121				125				130				135				140				145				150
VSPβ	L	S	L	G	F	K	I	V	F	L	S	G	R	Y	L	D	K	M	A	V	T	E	A	N	L	K	K	A	G	F
VSPβ-Met10	M						M											M						M						
VSPβ-Met20	M						M		M				M				M							M						M
VSPβ-Met30	M						M		M				M				M			M				M						M

	151				155				160				165				170				175				180
VSPβ	H	T	W	E	Q	L	I	L	K	D	P	H	L	I	T	P	N	A	L	S	Y	K	S	A	M	R	E	N	L	L
VSPβ-Met10																									M		M			
VSPβ-Met20													M				M				M				M		M			
VSPβ-Met30									M			M	M				M				M				M		M			

	181				185				190				195				200				205				210
VSPβ	R	Q	G	Y	R	I	V	G	I	I	G	D	Q	W	S	D	L	L	G	D	H	R	G	E	S	R	T	F	K	L
VSPβ-Met10									M													M								
VSPβ-Met20									M								M				M				M					
VSPβ-Met30									M								M				M									

	211			215		218
VSPβ	P	N	P	M	Y	Y	I	E
VSPβ-Met10			M		M			
VSPβ-Met20			M		M			
VSPβ-Met30			M		M			

FIG. 2.

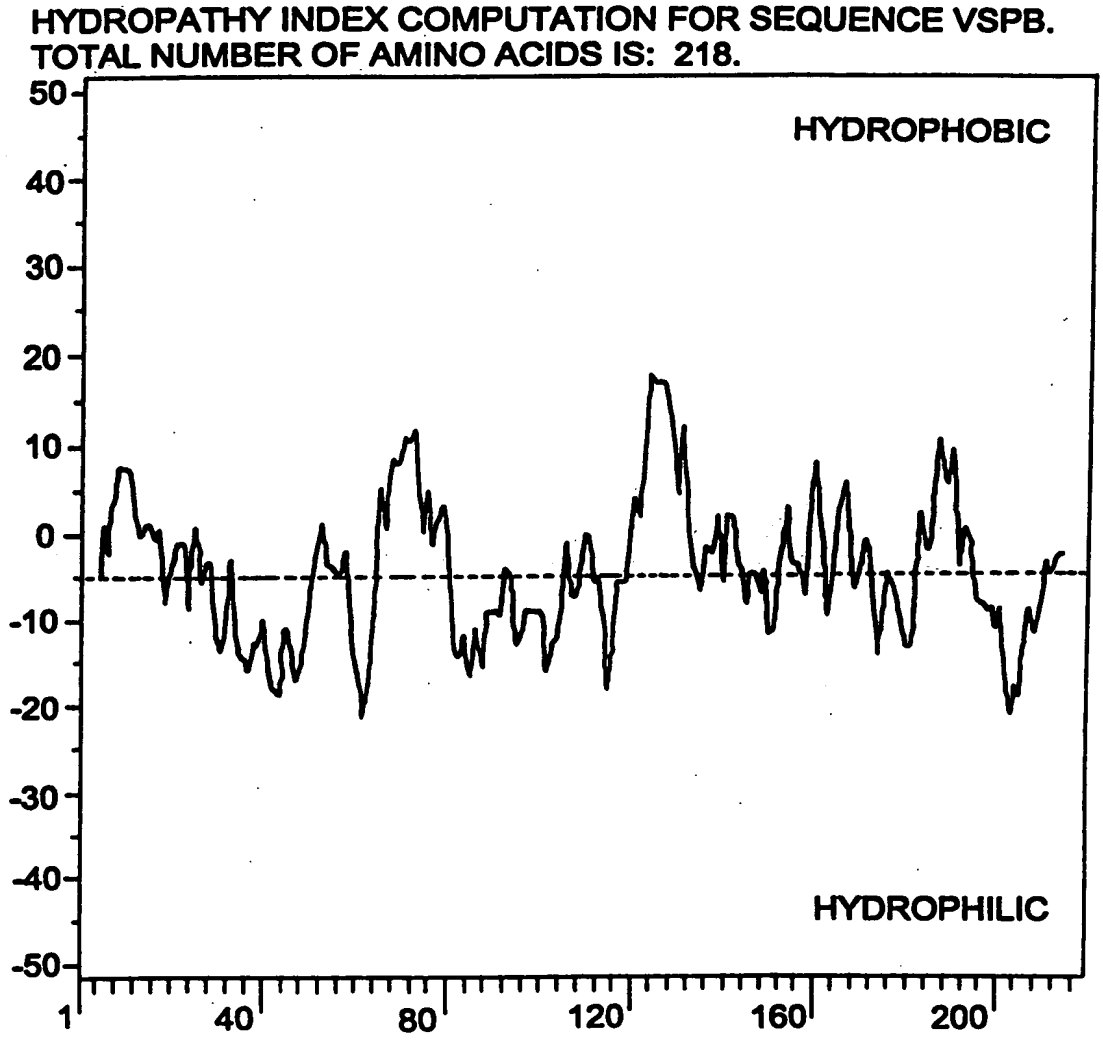

HYDROPATHY INDEX COMPUTATION FOR SEQUENCE VSPB.
TOTAL NUMBER OF AMINO ACIDS IS: 218.
HYDROPHOBIC
HYDROPHILIC
50
40
30
20
10
0
-10
-20
-30
-40
-50
1
40
80
120
160
200
HYDROPATHIC INDEX OF VSPB FROM AMINO ACID 1 TO AMINO ACID 218.
COMPUTED USING AN INTERVAL OF 9 AMINO ACIDS. (GRAVY=-4.95).

FIG. 3A.

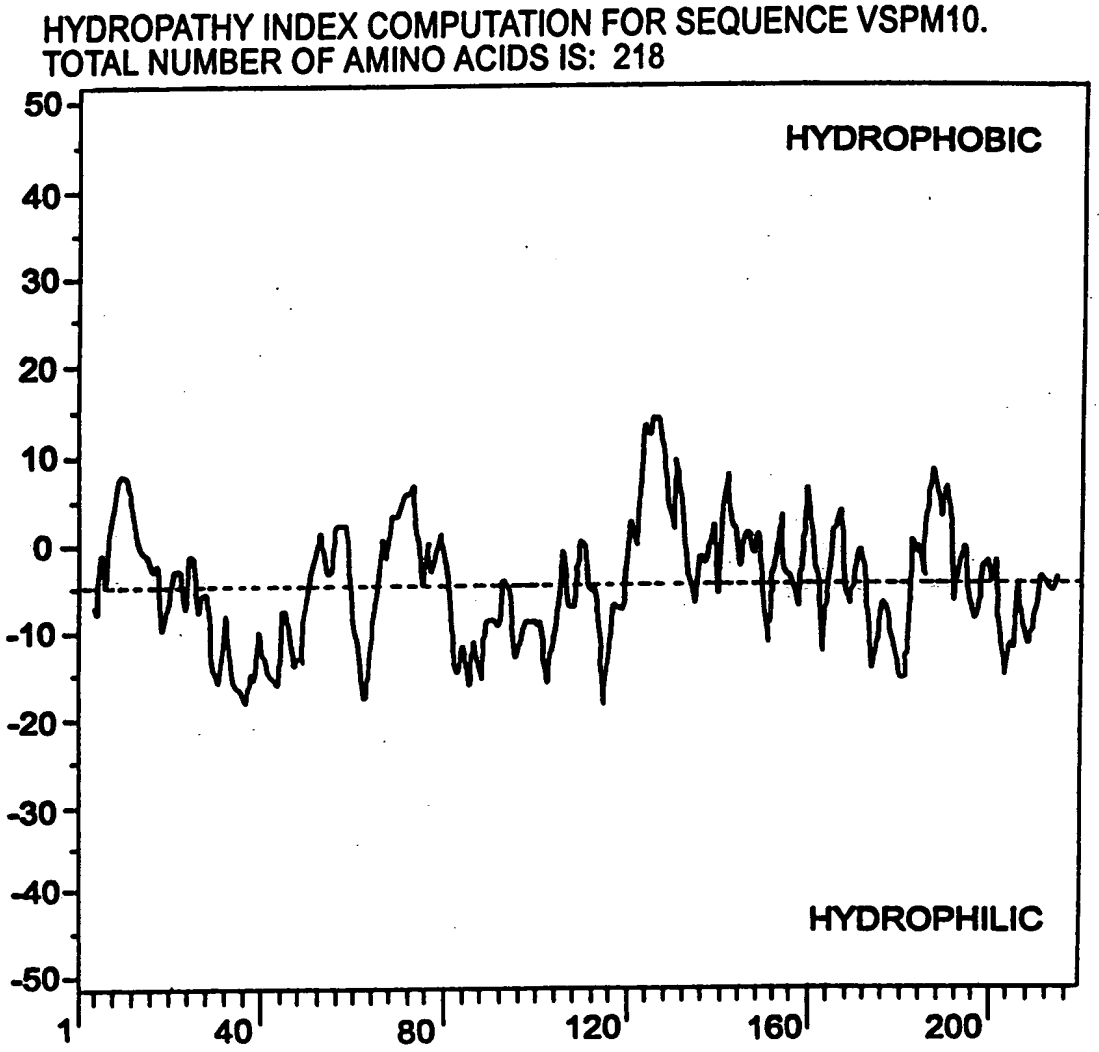

HYDROPATHY INDEX COMPUTATION FOR SEQUENCE VSPM10.
TOTAL NUMBER OF AMINO ACIDS IS: 218
HYDROPHOBIC
HYDROPHILIC
50
40
30
20
10
0
-10
-20
-30
-40
-50
1
40
80
120
160
200
HYDROPATHIC INDEX OF VSPM1 FROM AMINO ACID 1 TO AMINO ACID 218.
COMPUTED USING AN INTERVAL OF 9 AMINO ACIDS. (GRAVY=5.52).

FIG. 3B.

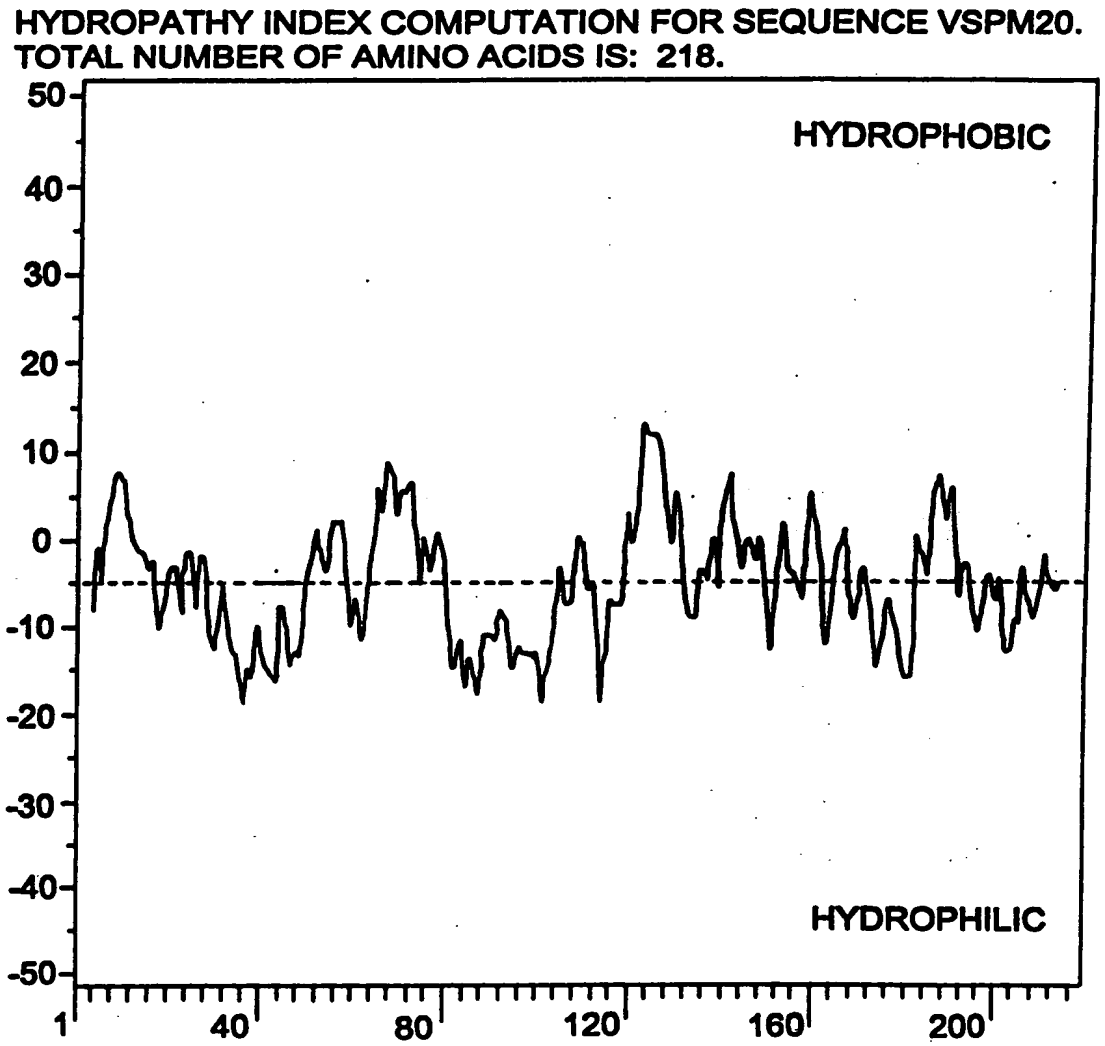

HYDROPATHY INDEX COMPUTATION FOR SEQUENCE VSPM20.
TOTAL NUMBER OF AMINO ACIDS IS: 218.
HYDROPHOBIC
HYDROPHILIC
50
40
30
20
10
0
-10
-20
-30
-40
-50
1
40
80
120
160
200
HYDROPATHIC INDEX OF VSPM20 FROM AMINO ACID 1 AMINO ACID 210.
COMPUTED USING AN INTERVAL OF 9 AMINO ACIDS. (GRAVY=-5.68).

FIG. 3C.

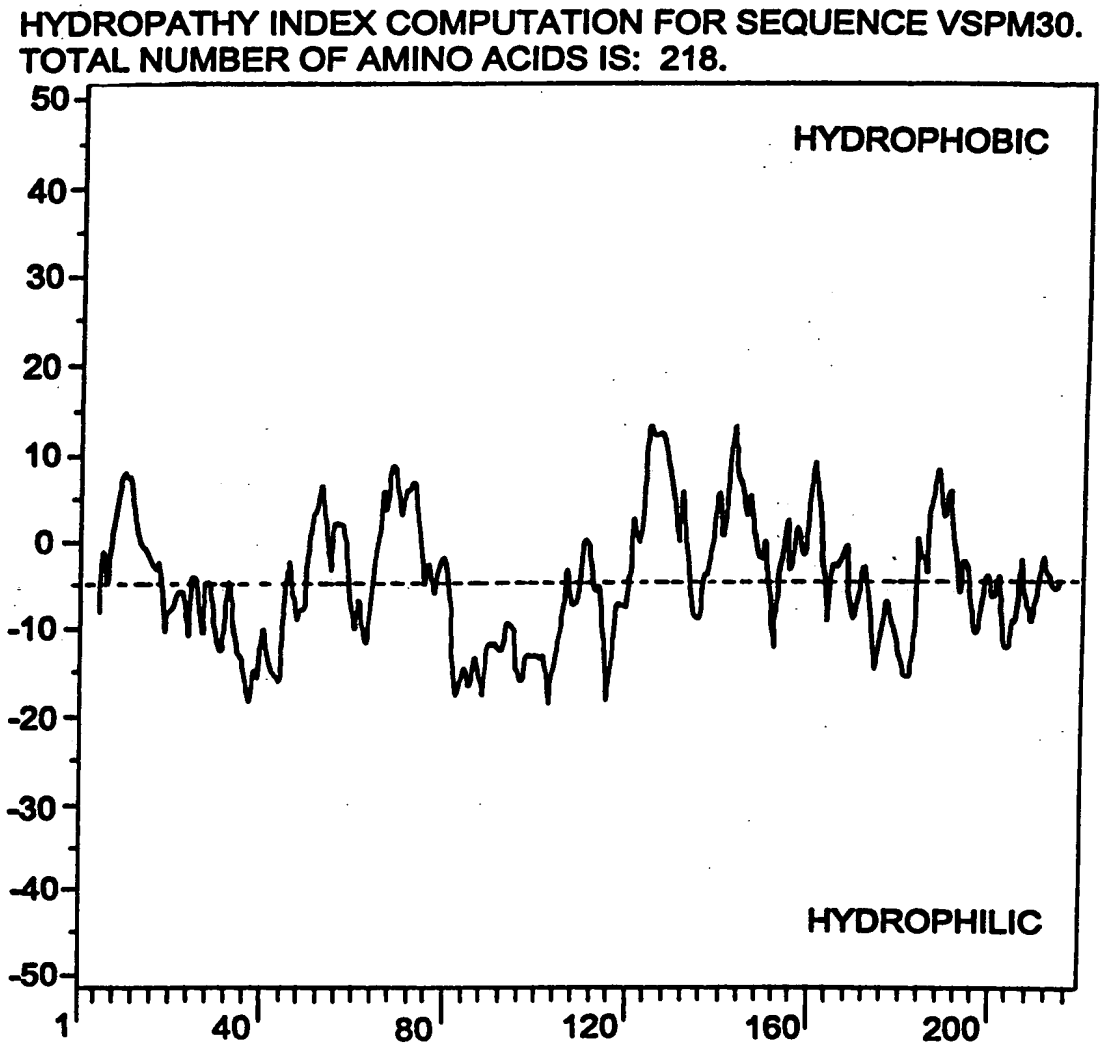

HYDROPATHY INDEX COMPUTATION FOR SEQUENCE VSPM30.
TOTAL NUMBER OF AMINO ACIDS IS: 218.
HYDROPHOBIC
HYDROPHILIC
50
40
30
20
10
0
-10
-20
-30
-40
-50
1
40
80
120
160
200
HYDROPATHIC INDEX OF VSPM30 FROM AMINO ACID 1 TO AMINO ACID 218.
COMPUTED USING AN INTERVAL OF 9 AMINO ACIDS. (GRAVY=-5.31).

FIG. 3D.

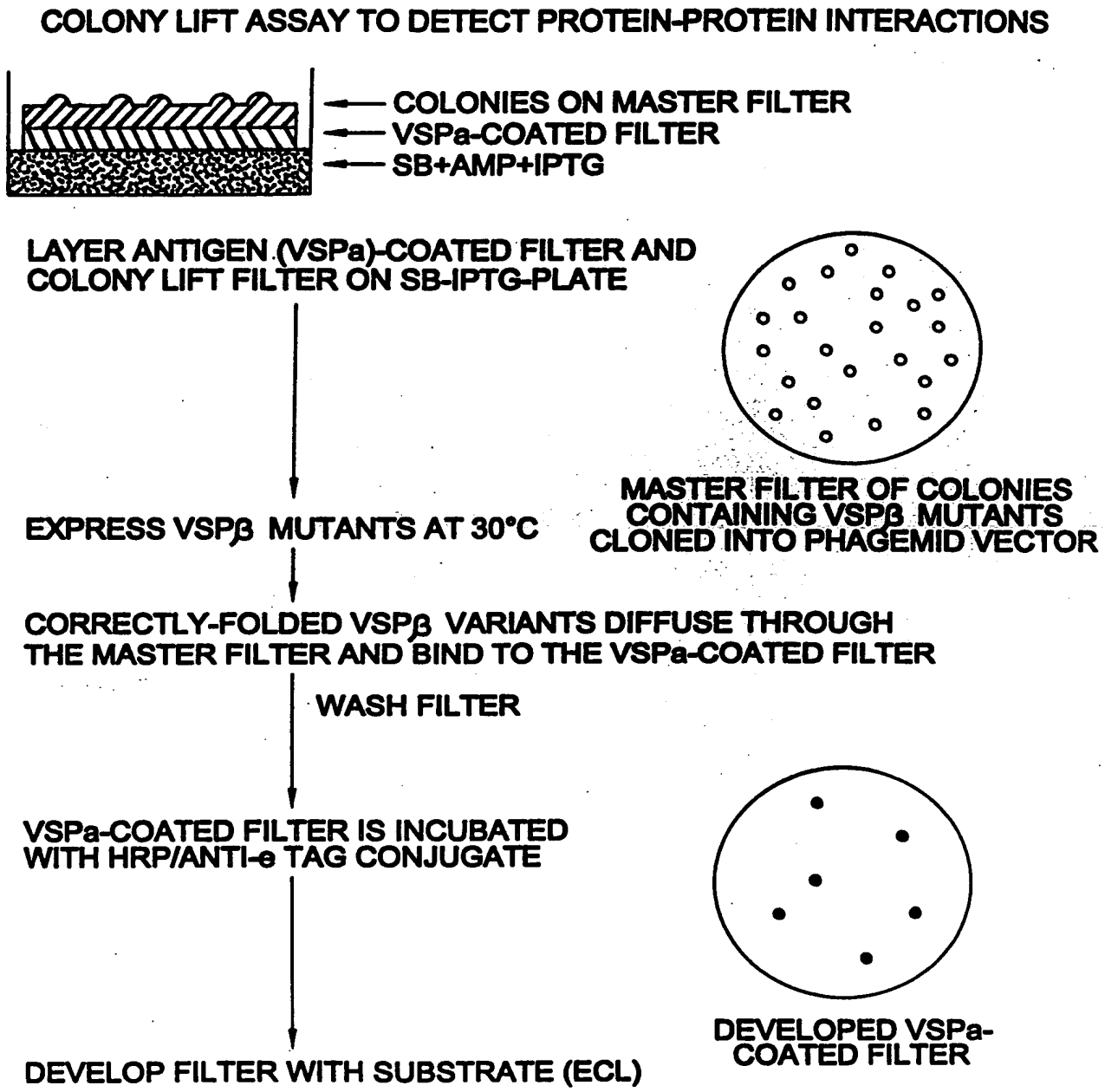

COLONY LIFT ASSAY TO DETECT PROTEIN-PROTEIN INTERACTIONS
COLONIES ON MASTER FILTER
VSPa-COATED FILTER
SB+AMP+IPTG
LAYER ANTIGEN (VSPa)-COATED FILTER AND COLONY LIFT FILTER ON SB-IPTG-PLATE
MASTER FILTER OF COLONIES CONTAINING VSPβ MUTANTS CLONED INTO PHAGEMID VECTOR
EXPRESS VSPβ MUTANTS AT 30°C
CORRECTLY-FOLDED VSPβ VARIANTS DIFFUSE THROUGH THE MASTER FILTER AND BIND TO THE VSPa-COATED FILTER
WASH FILTER
VSPa-COATED FILTER IS INCUBATED WITH HRP/ANTI-e TAG CONJUGATE
DEVELOPED VSPa-COATED FILTER
DEVELOP FILTER WITH SUBSTRATE (ECL)

FIG. 5.